Biodistribution of Gene Therapy Vectors for Phase 1 Clinical Trials

Anne M. Pilaro, Ph.D.

FDA/CBER

NIH Recombinant DNA Advisory Committee Meeting

Bethesda, MD

June 14, 1999

Initial Steps in the Development of a New Gene Therapy Agent

- **Product characterization**
  - manufacturing and quality control issues
- **Biologic activity**
  - *in vitro* and/or *in vivo* “proof of concept”
- **Safety**
  - toxicology testing in animals
    - safety
    - biodistribution

Guidance Documents for Preclinical Assessment of Therapeutic Products

- ICH M3 - Guidance on Nonclinical Safety Studies for the Conduct of Human Trials for Pharmaceuticals
  - provides guidance for the timing of conduct of animal studies in relationship to product development
  - recommends international standards for studies in support of marketing approval
  - does not specifically address gene therapy, but many of the approaches apply
- ***Fed Reg*, 62 (227): 62922, November 18, 1997**

Definition of Biodistribution (ICH M3)

- absorption, distribution, metabolism and excretion (ADME) of a drug or biologic
  - exposure data in animals should be evaluated prior to human clinical trials
  - ADME data in animals should be available to compare to human data in clinical trials
  - appropriate information (animal and human) should be available by completion of phase 1

Definition of Biodistribution Studies

- preclinical animal studies designed to determine distribution of vector to sites other than intended therapeutic site
  - read-out is presence of vector sequence by DNA PCR

Goals of Biodistribution Studies

designed to address two issues:

- dissemination of vector to the germline
  - total gonadal tissue assayed to date
- distribution of vector to non-target tissues
  - provides information on potential target organs for toxicity
- both issues may be addressed in same preclinical study

Results of the March 12, 1999 RAC Discussion on Gonadal Distribution

- open, scientific discussion of reproductive physiology, gonadal biodistribution results, and potential risks of positive signal to future generations
- risk of foreign gene transfer to germ cells, future progeny perceived low
  - acceptance by those present in context of somatic cell gene therapies

Results of the March 12, 1999 RAC Discussion on Gonadal Distribution

- biodistribution to gonads not necessarily needed prior to all phase 1 clinical trials
  - positive results not necessarily “show-stoppers”
  - negative results do not necessarily ensure “no risk”
- address lack of data, unknown risk(s) in consent form

Where Biodistribution Studies May be "Postponed"

- "previously defined" vector
  - previous experience with similar vector, route of administration, formulation, and schedule
    - *e.g.* adenovirus type 5 vectors
- transgene product "innocuous" if expressed ectopically
- size of vector (*i.e.* pDNA) not excessively different

And Where They May Not…

- new class of vector, no/little experience
  - *e.g.* AAV, lentivirus, others
- change in formulation (*i.e.* lipid carrier)
- change to intentional systemic route of administration with established vector
- transgene has the potential to induce toxicity if aberrantly expressed in non-target organ

Considerations in Designing Preclinical Biodistribution Studies

- **species selection**
  - non-human primates not always needed
- **animal gender**
  - male and/or female (driven by patient population)
- **animal number**
  - 3-5/sex/group minimum
    - use of smaller animals (i.e. rodents) allows determination in larger numbers

Considerations in Designing Preclinical Biodistribution Studies

- **dose selection**
  - vehicle control, maximally feasible/clinically relevant
  - lower dose for determination of NOEL desirable
- **intended clinical route of administration**
  - "worst-case" scenario may not adequately represent risk

Considerations in Designing Preclinical Biodistribution Studies

- **sacrifice time points**
  - goal is to determine kinetics of vector transduction, persistence
    - early - at time of peak vector transduction/expression
    - later - to be determined by intended clinical use
    - later still - to determine clearance of signal from gonads, non-target organs

Considerations in Designing Preclinical Biodistribution Studies

- **tissue panel for harvest**
  - minimum - peripheral blood, gonads, injection site
  - high-perfused organs (determination of toxicity)
    - brain, liver, lung, kidneys, heart, spleen, others
  - other tissues (e.g. draining, contralateral LN)
    - (un)expected target organs (determined by transgene)
- harvest tissues using clean instruments between organs, individual animals to minimize contamination

Considerations in Designing Preclinical Biodistribution Studies

- **detection assay**
  - methodology should detect sequence of vector DNA unique to that product
  - methodology should be appropriate to adequately detect vector sequence
    - in tissue samples from preclinical animal studies
    - in clinical samples obtained during the initial trial(s)

Example of Where "Generic" Biodistribution Studies May Be Useful

- one "giant" study to address everything
- distribution to non-target organs
  - after various routes of administration
  - after systemic administration
    - relative risk ratios
- effects of formulation and/or purification methodology change

Example of Where "Generic" Biodistribution Studies May Be Useful

- distribution to gonadal tissues
  - may be incorporated into non-target organ study
- results for specific class of vectors may be used to support other vectors in that class, regardless of transgene
  - if transgene has potential for toxicity, further studies may be needed

Summary

- biodistribution studies are designed to evaluate vector dissemination out of injected site
  - not only gonadal, but other non-target tissues
  - DNA PCR current accepted method
- studies not always required prior to phase 1
  - “previously defined vectors”, clinical context
  - will require data during course of product development

Sample Regulatory Letter for Studies Without Biodistribution Data

GONADAL BIODISTRIBUTION STUDIES:

"The present submission does not contain data that demonstrate the extent to which this vector is able to disseminate out of the injected site and distribute to gonadal tissues. These data are necessary to determine the risk of inadvertent gene transfer to the germ cells, which may result in genetic changes in subsequent progeny.

In the course of development of your product, you will be required to obtain these data and provide them to the Agency for review and comment. Data may be obtained either from biodistribution studies in animals, analysis of clinical samples, or from a combination of preclinical and clinical sample analyses. Clinical data should be derived from peripheral blood cells and semen samples during the treatment and follow-up periods for the clinical trial, and from gonadal tissues (primarily ova) obtained at autopsy from consenting patients. We will require that these data be provided in a timely fashion, so that the results may be used to guide further development and optimization of your product as a therapeutic agent.

Please update the Agency on the status of these studies at the time of each annual report."